Published in final edited form as:

Int J STD AIDS. 2017 April ; 28(5): 467–475. doi:10.1177/0956462416653001.

Impact of targeted counseling on reported vaginal hygiene practices and bacterial vaginosis: the HIV Prevention Trials Network 035 study

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Abstract

Objective—We describe the impact of intense counseling to reduce vaginal hygiene practices and its effect on bacterial vaginosis (BV).

Design—Secondary data analysis of HPTN 035 trial

Setting—Seven African and one U.S. site

Population—HIV negative, non-pregnant women at least 18years old

Methods—At enrollment and during follow-up quarterly visits, vaginal hygiene practices were determined by face-to-face administration of a behavioral assessment questionnaire. Vaginal hygiene practices were categorized as insertion into the vagina of: (1) nothing; (2) water only; and (3) other substances with or without water. Each practice was quantified by frequency and type/combination of inserted substances. At quarterly visits, diagnosis of BV was made using the

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Nugent score. Trends for vaginal hygiene practices and BV were evaluated using generalized estimating equation models.

Results—3087 participants from the HPTN 035 study were eligible for this analysis. At enrollment, 1859 (60%) reported recent vaginal hygiene practices. By one year, this figure had decreased to 1019 (33%) with counseling. However, BV prevalence remained consistent across the study observation period, with 36–38% of women testing positive for the condition (p for trend= 0.27). Overall, those who reported douching with water only (AOR= 1.03, 95%CI: 0.94 – 1.13) and those who reported inserting other substances (AOR= 0.98, 95%CI: 0.88 – 1.09) in the past quarter were not more likely to have BV compared to those who reported no insertions. However, in South Africa, an increase in BV was seen among those who reported inserting other substances (AOR: 1.48, 95%CI: 1.17, 1.88).

Conclusions—Targeted counseling against vaginal hygiene practices resulted in change in self-reported behavior, but did not have an impact on BV diagnosis in all but one site.

Keywords

Vaginal hygiene practices; Counselling; Bacterial vaginosis

Introduction

Vaginal hygiene practices are genital cleansing practices that women engage in to address perceived woman-specific needs of hygiene, sexuality, and vaginal health and treatment of genital symptoms.[1–3] These behaviors, including physical or chemical manipulation of vaginal tissue by application of soaps, acetic acid, douches, or insertion of natural or synthetic fibers, or other efforts to change the normal environment of the vagina may result in changes to pH, vaginal dryness, disruption of bacterial flora, or micro-abrasions or erosions. These complications increase a woman's susceptibility for infection, such as bacterial vaginosis (BV) and sexually transmitted infections including HIV, as well as other adverse gynecologic and obstetric outcomes.[2, 4–11] The prevalence of vaginal hygiene practices is believed to be high, although estimates range from 6 to 98% in sub-Saharan Africa.[1]

A consistent association has been observed between certain vaginal hygiene practices and BV among women.[4, 12, 13] Ness *et al.* found that douching was associated with twice the frequency of BV (OR 2.1; 95% CI 1.3–3.1), with the highest risk among women reporting so in the past week.[12] Other studies have shown a high correlation between vaginal hygiene practices, including douching with soap, antiseptics, commercial douches, homemade solutions, and cleansing post menses as well as application of lubricants such as petrolatum or saliva, and BV.[8,14,15]

During HIV Prevention Trial Network protocol 035 (HPTN 035), a multi-center study that evaluated the safety and effectiveness of two candidate microbicide gels BufferGel and PRO 2000 0.5% gel, we conducted intensive counseling to inform women of the potential unhealthy consequences of vaginal hygiene practices.[16] Early studies of BufferGel and PRO 2000 0.5% gel indicated that these agents were not associated with development of BV.[14,17]

In this secondary analysis, we investigated the impact of one-on-one counseling on self-reported vaginal hygiene practices and BV. We hypothesized that a positive correlation would exist between vaginal practices and prevalent BV.

Methods

The design of HPTN 035 has been described previously.[16] Briefly, we conducted the study between February 2005 and October 2008. We enrolled HIV negative, non-pregnant women at least 18 years old and randomized them to one of four study arms: BufferGel vaginal gel, PRO 2000 0.5% vaginal gel, hydroxyethylcellulose (HEC) placebo vaginal gel, or no gel/condoms only. Participants were enrolled for a minimum of 12 months, the timing of the study's primary outcome, but follow-up continued until the last participant reached this time point for a maximum of 30 months. The study was conducted in seven African (Blantyre, Malawi; Lilongwe, Malawi; Durban, South Africa; Hlabisa, South Africa; Kamwala, Zambia; Chitungwiza/Harare, Zimbabwe) and one U.S. site (Philadelphia, Pennsylvania). The trial (clinicaltrials.gov number NCT00074425) was approved and overseen by local ethics committees and institutional review boards at each study site. All participants provided written informed consent prior to participation.

At enrollment and during quarterly visits, we administered a behavioral assessment questionnaire to all participants, which included a detailed question regarding vaginal practices (Figure 1). This interview was conducted in a private setting by a trained research staff member, generally a nurse or other clinician, and was followed by a physical examination that included collection of a vaginal fluid for pH, Gram's stain, and wet mount. All sites collected vaginal smears for central processing and adhered to HPTN Central Laboratory standards for collection, processing, labelling and transportation.[16]

At a later time during the same visit, participants were counseled on HIV/STI risk reduction and against vaginal hygiene practices. This conversation aimed to communicate several principals in regards to vaginal hygiene practices: (i) vaginal hygiene practices, especially those affecting lubrication of vaginal mucosa, could put the individual at increased risk for contracting HIV/AIDS, (ii) since the study utilizes vaginally applied products, it is important to avoid co-application of other products, (iii) vaginal hygiene practices can disrupt vaginal pH balance and put the woman at risk for vaginal infections, and (iv) avoiding vaginal hygiene practices promotes vaginal health. Specific messages were catered to an individual based on her responses to the questionnaire and her specific practices. This non-scripted counseling occurred at every contact (including unscheduled interim visits), was one-on-one with a nurse and/or physician, and was followed by a verbal assessment of participant understanding.

Our statistical analysis was designed to describe vaginal hygiene practices over time and evaluate their relationship to the diagnosis of BV. We included all follow-up data from participants, including observations past 12 months in participants enrolled for longer periods. Vaginal practices were categorized as follows: (1) Did not insert anything into the vagina; (2) insertion of water only; and (3) insertion of any other substances with or without water (e.g., water with vinegar, water with soap, paper, cloth, cotton or cotton wool,

tampons, fingers without anything else). For the last two categories, follow-on questions were asked to delineate frequency and type/combination of inserted substance. Diagnosis of BV was based on the Nugent criteria.[17] The validation of the vaginal Gram stain smear compared with the Amsel criteria found the sensitivity and specificity was 89 and 83%, respectively and hence the vaginal Gram stain (Nugent criteria) was considered to be a sensitive method for the diagnosis of bacterial vaginosis.[18] Presence of BV was defined as a Nugent score of 7–10 and absence of BV was defined by a Nugent score of 0–6. Symptomatic women diagnosed with BV (Nugent ≥7 were provided 2 grams of metronidazole as a single dose for treatment at the same visit as recommended.[19] All eligible, enrolled and randomized participants in the HPTN 035 study were included in the analysis.

Baseline characteristics of participants were compared across the clinical sites using the Chi-square test for independence for categorical variables, and analysis of variance for continuous measures. Nugent scores for BV diagnoses were represented graphically over time. We also examined the association between vaginal hygiene practices and BV positivity overall and by country using generalized estimating equations (GEE) with a logit link function and exchangeable working correlation structure. We ran all models, we adjusted for country/sites, arm assignment (BufferGel, no gel, PRO 2000/5, placebo), quarterly visit beginning with the first quarterly visit at 3 months through the study exit visit at 32 months, and potential confounders of age (in 10 year increments), education (some primary school or less, secondary school or more), marital status (married, has a partner or neither) and number of sex partners (0, 1, 2, 3 or more).

All analyses were performed using SAS Version 9.2 (SAS Institute, Cary, North Carolina, USA).

Results

Across the eight enrolling sites, 3,087 participants were included in the analysis. The median follow-up time was 20.4 months. The mean age of participants was 26.3 years (SD 6.2); educational status varied across countries. Nearly all participants reported being married and 86.7% reported partner/husband providing financial support (Table 1). For all characteristics, statistically significant differences were observed across the countries ($p<0.0001$).

At baseline, out of 3087 participants, 1228 (40%) denied vaginal hygiene practices, while 424 (14%) reported using water, and 1435 (46%) reported using various other products including commercial douches, vinegar and water, soap and water, paper, cloth, cotton, cotton wool, tampons, or fingers. The percent of participants using various vaginal hygiene practices is shown over time in Figure 2. Women who reported not practicing any form of vaginal hygiene practice increased from 40% at enrollment to 67% by 12 months ($p<0.0001$); this upward trend appeared to wane during the second year. General downward trends were observed for those reporting using water ($p<0.0001$), and other substances during the first year ($p<0.0001$).

Alongside the self-reported behavioral data, we examined trends in BV over time as a biological marker influenced by vaginal hygiene practices. While frequency of BV testing waned after one year as patients exited the study, the proportion testing positive for BV remained constant (approximately 36–38%) throughout the follow-up period (Figure 3 and Table 2).

Overall, compared to insertion of nothing, we observed no statistically significant effects of water use only (AOR: 0.98; 95% CI: 0.88, 1.09; p=0.75) or use of materials or products other than or in addition to water (AOR: 1.04; 95% CI: 0.95, 1.13; p=0.44) on BV positivity after adjusting for study visit, arm assignment, age, education, marital status and number of sex partners (Table 3 and Figure 4). Compared to insertion of nothing, the effect of water use only and use of materials or products other than or in addition to water on BV positivity, respectively, by country were as follows: Malawi: (AOR: 0.89; 95% CI: 0.78, 1.03; p=0.12) and (AOR: 0.91; 95% CI: 0.80, 1.03; p=0.13); South Africa: (AOR: 0.78; 95% CI: 0.22, 2.84; p=0.72) and (AOR: 1.48; 95% CI: 1.17, 1.88; p=0.001); U.S.A: (AOR: 0.86; 95% CI: 0.38, 1.96; p=0.73) and (AOR: 1.16; 95% CI: 0.91, 1.48; p=0.23); Zambia: (AOR: 1.10; 95% CI: 0.87, 1.38; p=0.43) and (AOR: 0.98; 95% CI: 0.72, 1.32; p=0.88); Zimbabwe: (AOR: 0.86; 95% CI: 0.62, 1.20; p=0.38) and (AOR: 0.76; 95% CI: 0.39, 1.51; p=0.44). To account for multiple comparisons [i.e., Two overall comparisons and two comparisons each for five countries (12 total)], we used a Bonferroni adjusted p-value of $p < 0.004$ to attribute meaningful statistical significance at the $p < 0.05$ nominal level. Therefore, in South Africa only, we observed increased odds of BV positivity with use of materials or products other than or in addition to water compared to insertion of nothing.

Discussion

In HPTN 035, self-reported vaginal hygiene practices declined among study participants over time. This was likely a result of the counseling provided by the study, which engaged and educated participants regarding the risks associated with vaginal hygiene practices. Interestingly, the analogous trends for BV diagnosis – our proposed surrogate marker for vaginal hygiene practices – did not align with the change in reported behavior.

This analysis possesses numerous strengths: conduct within a large, rigorous clinical trial and a generalizable population with high prevalence of studied behaviors. Nevertheless, we acknowledge limitations as well. First, the intervention was designed as a multifaceted counseling session and covered many topics on health. Due to time constraints, messages were covered generically but with latitude as to the “dose” of messages for each particular participant. While it is probable that every participant received counseling against vaginal hygiene practices at every visit, there may have been variability in the amount of time helping each participant understand the reasons for changing behavior or to discuss alternatives and skills to help individuals change their behavior. This could have resulted in a superficial understanding on part of the participant, which may have resulted in a reduction but not cessation of high-risk practices. Second, while the analysis is very robust in the first 12–15 months, participation in study declined as participants exited study in the second year. This could affect our ability to distinguish changes in trends during the latter follow-up period. Finally, we do not have data available to determine which cases of BV were treated,

i.e., which were symptomatic, to know if subsequent cases of BV were incident cases or whether they were persistent BV from previous episodes. Treatment was only for symptomatic cases; therefore, the previously diagnosed asymptomatic cases could potentially contribute to the BV cases at the next assessment. This could have diluted the effect of positive behavior change that did occur, as untreated BV acquired during previous time periods (i.e. prior behavior) would be attributed falsely during later time points when counseling could have been having its impact. This would have influenced the result toward the null hypothesis, that there was no impact of change in vaginal hygiene practices on BV acquisition, when in fact change did occur, and observed subsequent BV was prevalent, not incident. Furthermore, the relapse rate of BV in these circumstances is approximately 20 to 50% following treatment, which would occur regardless of ongoing behavior change in regards to vaginal hygiene practice.[20]

Other studies have found similar results. Most were smaller and vulnerable to biases resulting from self-report – as in the current study – but they did lead to the development of an individualized counseling approach acknowledging the perceived benefits and risks of vaginal hygiene practices. Notably, Masese *et al.* used an information-motivation framework including individualized skills teaching and harm reduction to encourage participants to reduce or eliminate vaginal washing. They found a reduction in self-report of vaginal hygiene practices but no significant changes on biological markers, including prevalence of BV.[20] Similarly, other groups have shown impact of individualized counseling on self-reported behaviors but no conclusive evidence that those changes in behavior led to reductions in BV.[3, 21, 22] Our discordant findings suggest that either the link identified between vaginal hygiene practices and BV is not causal or, self-reported behavior change is an unreliable method to assess changes in this behavior. We did note one exception to this overall finding. When we conducted stratified analysis of South African data, we found that counseling around vaginal practices – in particular, products other than or in addition to water – was associated with BV. This result is in line with our original hypothesis and reiterates the potential for heterogeneity between populations. It is unlikely that retention or adherence to gel could have caused this difference as both were high and similar at all sites.[16]

Self-reported behavior change in our study could have been influenced by social desirability bias. Data on vaginal hygiene practices was collected through face-to-face interviews. While this is an effective means for counseling, data collection could have been more reliable if ascertained through less personal means, such Audio Computer Assisted Self Interview (ACASI). ACASI was not used in this study. It has been known to capture some sensitive behaviors more reliably in high- risk populations and standardizes data collection.[23,24] While an over-estimation of actual change in vaginal hygiene practices could partially explain the result, it is also possible that our expectation that compliance to the counseling would be cessation of vaginal hygiene practices altogether was not the interpretation of the participants. Perhaps this explains why there are some who changed their behavior only in terms of frequency or type of inserts. Despite its association with our exposure of interest, it is also possible that BV may not be the ideal biological marker to assess changes in vaginal hygiene practices.

Taken together, these findings highlight some of the challenges involved with evaluating change within behavioral interventions, especially those of a sensitive nature in which there are currently no reliable biological proxies to measure. These findings advocate for use of more rigorous ways to evaluate personal behaviors and in the development of better objective and biological markers for monitoring these behaviors.[24]

Conclusions

Intense and continuous counseling against vaginal hygiene practices such as occurred in the HPTN 035 trial was associated with improvements in self-reported behaviors but the trends of BV were not affected, even in the context of a rigorously conducted clinical trial. These data illustrate the potential difficulties in modifying and measuring behavior within the realm of reproductive health. However, this study has demonstrated that nesting these types of behavioral interventions within larger trials, such as HIV prevention trials, may provide an avenue with sufficient statistical power to answer these questions in the future. Outcomes of such may inform the design of public health strategies to reduce behavioral practices potentially harmful to health.

Supplementary Material

Refer to Web version on PubMed Central for supplementary material.

Acknowledgments

We sincerely thank the women who participated in this study. We gratefully acknowledge the HPTN 035 study team and study sites for their work on data and sample collection and the Statistical Center for their work on data management

Funding

HIV Prevention Trials Network (HPTN) 035 was funded by the US National Institutes of Health (NIH). The HPTN (U01AI46749) has been funded by the National Institute of Allergy and Infectious Diseases (NIAID), the Eunice Kennedy Shriver National Institute of Child Health and Human Development (NICHD), National Institute of Drug Abuse (NIDA), and National Institute of Mental Health (NIMH). The Microbicide Trials Network (U01AI068633), which implemented HPTN 035, was funded by NIAID, NICHD, and NIMH. Additional trainee support was provided by the Vanderbilt-CIDRZ AIDS International Training and Research Program (D43TW001035).

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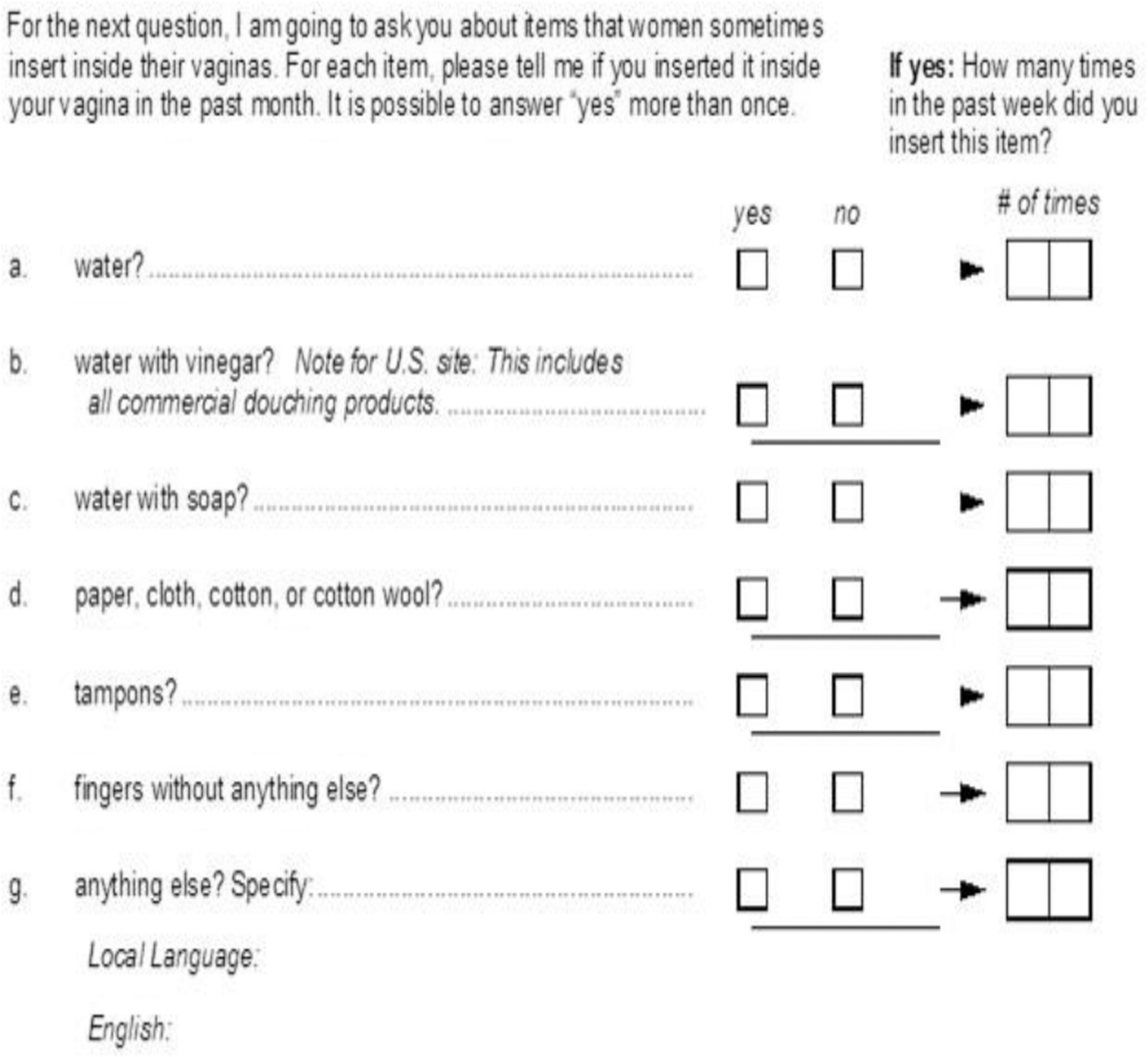

Figure 1.
Vaginal hygiene practice questions

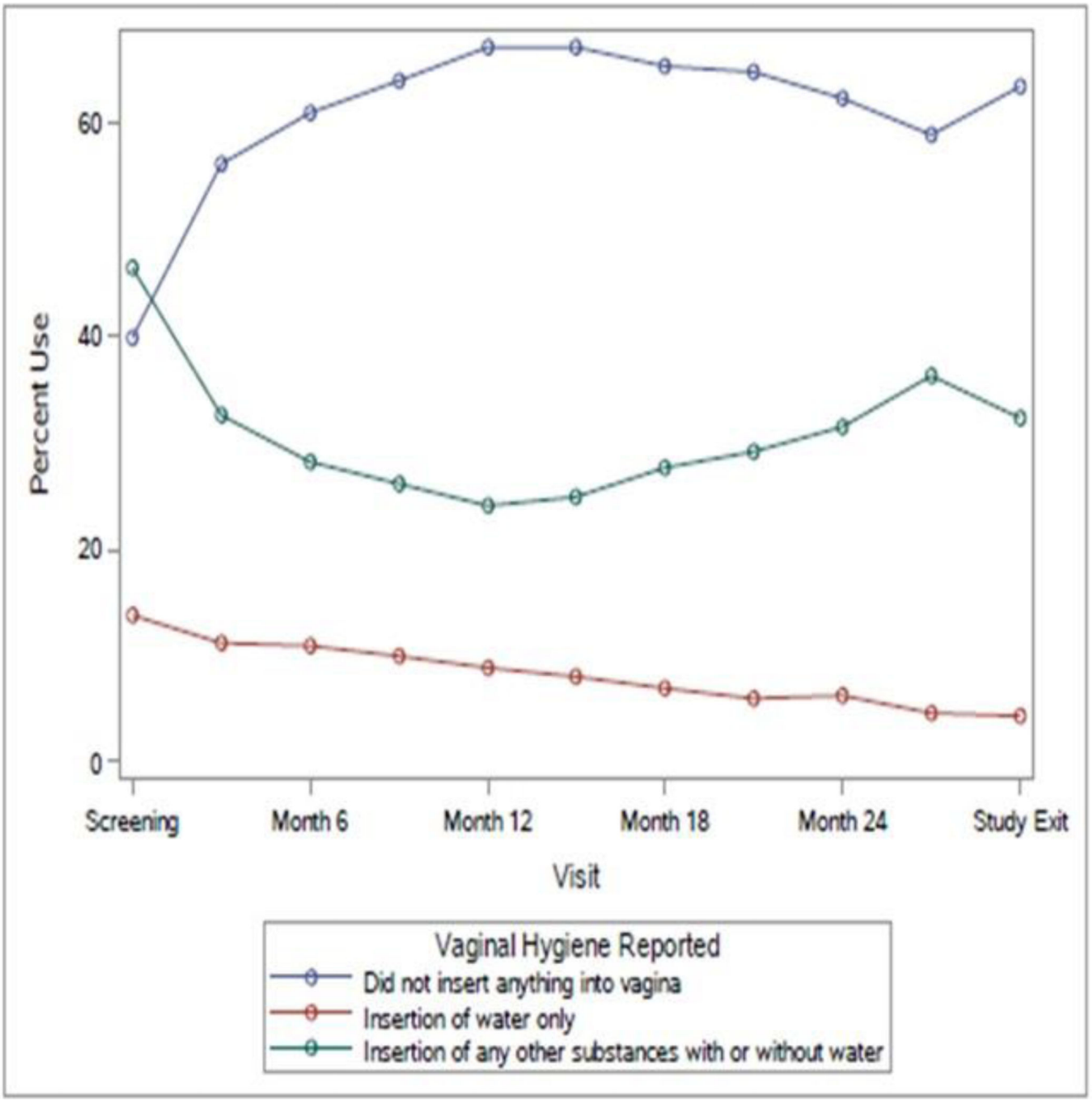

Figure 2.
Vaginal hygiene practices reported in last month by study visit

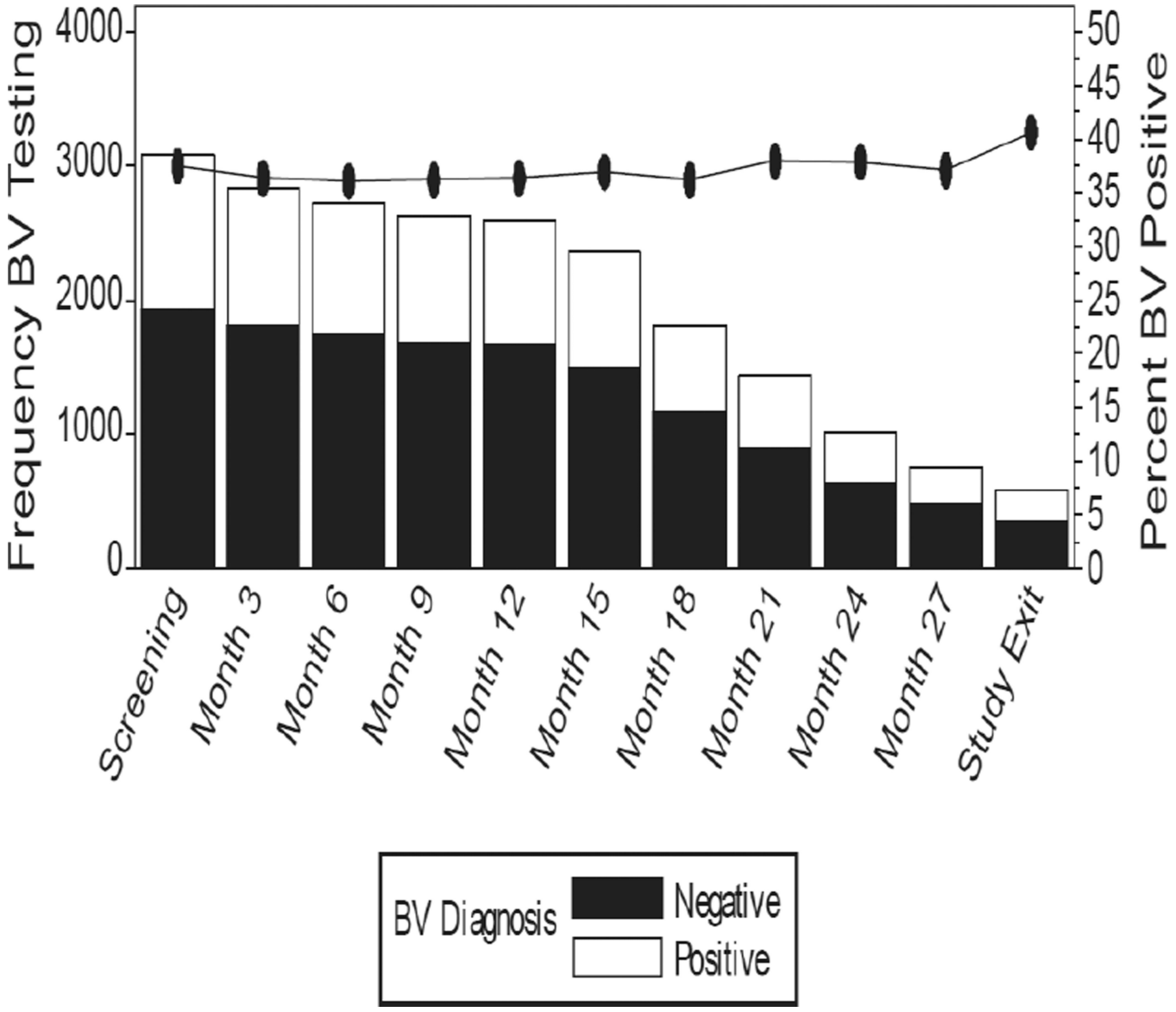

Figure 3.
Distribution of bacterial vaginosis according to study visit

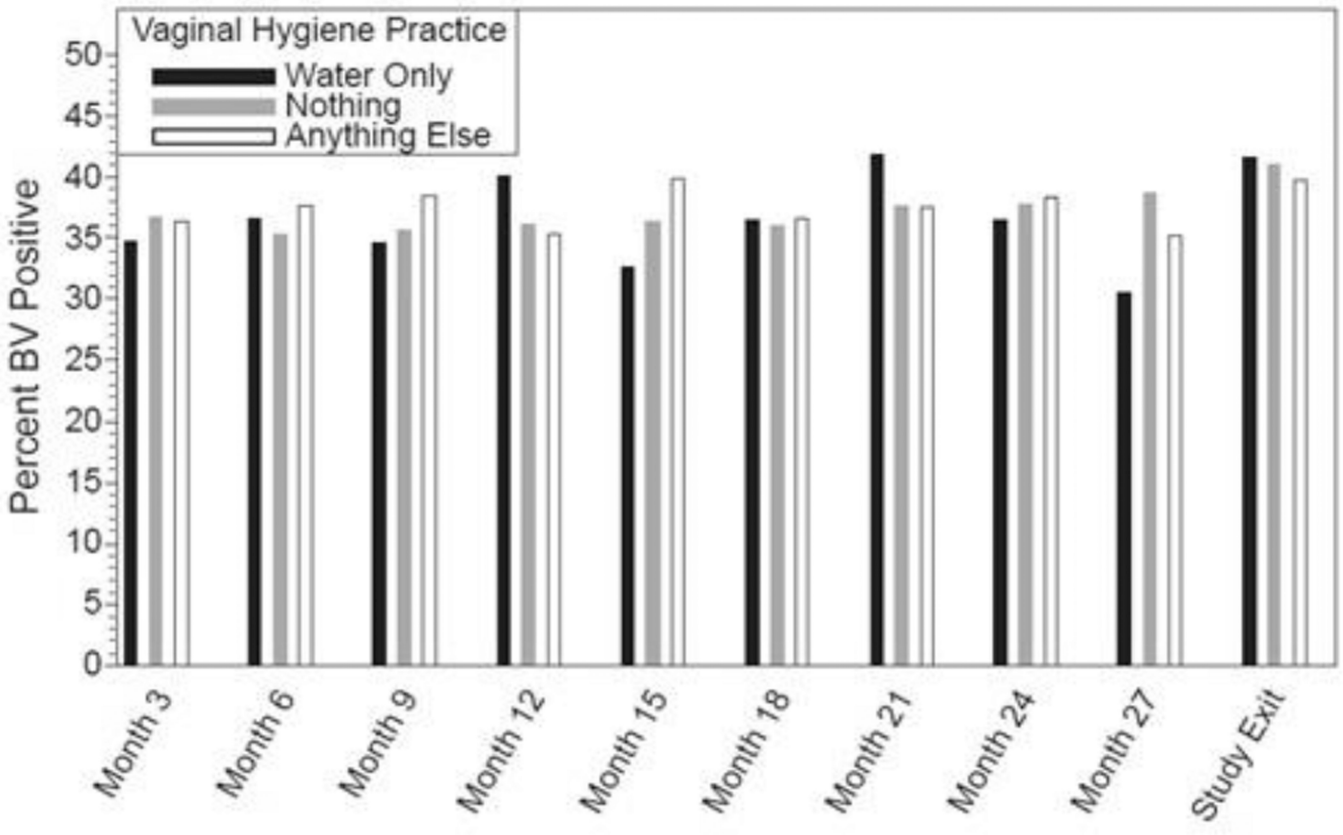

Figure 4.
Proportion of women with bacterial vaginosis over time according to reported vaginal hygiene practice

Int J STD AIDS. Author manuscript; available in PMC 2017 April 01.

Table 1

Baseline social and demographic characteristics of women enrolled in the HPTN 035 study by site

Site / Variable[1,2]	Malawi (n=1037)	South Africa (n=1048)	United States (n=200)	Zambia (n=319)	Zimbabwe (n=483)	Total (n=3087)
Age	26.7 (5.9)	25.2 (5.0)	35.5 (9.9)	23.0 (3.5)	26.3 (4.5)	26.3 (6.2)
Education						
Primary School or Less	805 (77.7)	123 (11.7)	3 (1.5)	179 (56.1)	32 (6.6)	1142 (37.0)
Secondary School or More	231 (22.3)	925 (88.3)	197 (98.5)	140 (43.9)	451 (93.4)	1944 (63.0)
Marital Status						
Husband	1013 (97.7)	161 (15.4)	28 (14.0)	258 (80.9)	466 (96.5)	1926 (62.4)
Partner	24 (2.3)	885 (84.5)	170 (85.0)	61 (19.1)	17 (3.5)	1157 (37.5)
Neither	0 (0.0)	2 (0.2)	2 (1.0)	0 (0.0)	0 (0.0)	4 (0.1)
Husband/Partner Provides Financial Support	998 (96.2)	748 (71.5)	150 (75.8)	305 (95.6)	472 (97.7)	2673 (86.7)
Partner Education						
Primary School or Less	554 (53.4)	105 (10.0)	4 (2.0)	50 (15.7)	23 (4.8)	736 (23.9)
Secondary School or More	442 (42.6)	895 (85.6)	183 (92.4)	262 (82.1)	451 (93.4)	2233 (72.4)
Don't Know	41 (4.0)	46 (4.4)	11 (5.6)	7 (2.2)	9 (1.9)	114 (3.7)
Number of Current Sex Partners	1.0 (0.1)	1.0 (0.3)	1.5 (1.4)	1.0 (0.2)	1.0 (0.1)	1.1 (0.4)
Vaginal Practices						
Did Not Insert Anything into Vagina	128 (12.3)	845 (80.6)	39 (19.5)	72 (22.6)	144 (29.8)	1228 (39.8)
Insertion of Water Only	145 (14.0)	20 (1.9)	1 (0.5)	58 (18.2)	200 (41.4)	424 (13.7)
Insertion of Any Other Substances[3] with or without Water	764 (73.7)	183 (17.5)	160 (80.0)	189 (59.3)	139 (28.8)	1435 (46.5)

[1] All values are expressed as N (%), except age and number of current sex partners which are shown as the mean (standard deviation).

[2]Chi-square tests (for categorical variables) or ANOVA (for the age variable only) across study sites showed $p < 0.0001$ for all variables.

[3]This includes water with vinegar, water with soap, paper, cloth, cotton or cotton wool, tampons, fingers without anything else, and anything else.

Table 2

Distribution of Nugent scores by study visit

	Study Visit										
Nugent Score Category N (%)	**Screening (N=3078)**	**Month 3 (N=2831)**	**Month 6 (N=2725)**	**Month 9 (N=2619)**	**Month 12 (N=2600)**	**Month 15 (N=2362)**	**Month 18 (N=1809)**	**Month 21 (N=1439)**	**Month 24 (N=1013)**	**Month 27 (N=757)**	**Study Exit (N=588)**
0–3	1383 (44.9)	1384 (48.9)	1306 (47.9)	1246 (47.6)	1206 (46.4)	1108 (46.9)	851 (47.0)	659 (45.8)	459 (45.3)	332 (43.9)	257 (43.7)
4–6	540 (17.5)	417 (14.7)	435 (16.0)	424 (16.2)	448 (17.2)	381 (16.1)	303 (16.8)	234 (16.3)	170 (16.8)	144 (19.0)	92 (15.7)
7–10	1155 (37.5)	1030 (36.4)	984 (36.1)	949 (36.2)	946 (36.4)	873 (37.0)	655 (36.2)	546 (37.9)	384 (37.9)	281 (37.1)	239 (40.7)

Table 3

Adjusted odds ratios showing the effect of vaginal hygiene practices on bacterial vaginosis

	Overall	Malawi	South Africa	U.S.A.	Zambia	Zimbabwe
	Adjusted Odds Ratio (95% Confidence Interval)					
Vaginal hygiene practice						
Water only	0.98 (0.88, 1.09)	0.89 (0.78, 1.03)	0.78 (0.22, 2.84)	0.86 (0.38, 1.96)	1.10 (0.87, 1.38)	0.86 (0.62, 1.20)
Any other substance, with or without water	1.04 (0.95, 1.13)	0.91 (0.80, 1.03)	1.48 (1.17, 1.88)	1.16 (0.91, 1.48)	0.98 (0.72, 1.32)	0.76 (0.39, 1.51)
Did not insert anything into vagina	Ref	Ref	Ref	Ref	Ref	Ref
Arm Assignment						
BufferGel	0.96 (0.82, 1.12)	1.03 (0.79, 1.34)	0.89 (0.69, 1.16)	0.91 (0.52, 1.57)	0.95 (0.57, 1.57)	1.01 (0.67, 1.52)
No Gel	1.0 (0.86, 1.16)	1.07 (0.82, 1.39)	1.01 (0.79, 1.31)	0.69 (0.39, 1.21)	1.14 (0.70, 1.86)	0.87 (0.58, 1.30)
PRO 2000/5	1.02 (0.88, 1.19)	0.93 (0.72, 1.21)	1.15 (0.89, 1.47)	0.82 (0.46, 1.47)	1.29 (0.79, 2.11)	0.86 (0.58, 1.27)
Placebo	Ref	Ref	Ref	Ref	Ref	Ref
Study Quarter	1.01 (1.00, 1.03)	1.00 (0.98, 1.03)	1.02 (1.0, 1.05)	0.99 (0.95, 1.03)	1.08 (1.02, 1.15)	1.02 (0.97, 1.07)
Age (10 year increments)	1.07 (0.98, 1.16)	0.99 (0.85, 1.16)	1.15 (0.93, 1.41)	1.11 (0.91, 1.35)	1.45 (0.89, 2.37)	0.87 (0.63, 1.19)
Educational Status						

	Overall	Malawi	South Africa	U.S.A.	Zambia	Zimbabwe
	Adjusted Odds Ratio (95% Confidence Interval)					
Primary school or less	Ref	Ref	Ref	Ref	Ref	Ref
Secondary school or more	0.90 (0.80, 1.03)	0.81 (0.64, 1.02)	1.03 (0.76, 1.39)	1.32 (0.39, 4.44)	0.83 (0.58, 1.20)	1.09 (0.60, 1.98)
Marital Status						
Husband or neither	0.80 (0.71, 0.91)	0.50 (0.28, 0.90)	0.57 (0.43, 0.76)	0.81 (0.46, 1.43)	0.63 (0.40, 0.98)	0.61 (0.28, 1.35)
Partner	Ref	Ref	Ref	Ref	Ref	Ref
Number of Sex Partners						
0	3.63 (1.58, 8.33)	2.28 (1.85, 2.80)	5.95 (2.08, 17.03)	NA	NA	NA
1	Ref	Ref	Ref	Ref	Ref	Ref
2	1.28 (0.93, 1.76)	3.14 (2.28, 4.32)	1.94 (1.09, 3.45)	0.81 (0.48, 1.37)	0.74 (0.33, 1.63)	3.07 (1.90, 4.96)
3 or more	0.80 (0.44, 1.47)	NA	0.10 (0.03, 0.35)	0.91 (0.47, 1.78)	0.26 (0.14, 0.46)	NA